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**VITAMIN E REQUIREMENTS OF ADULT DOMESTIC
CATS (*FELIS CATUS*) FED DIETS CONTAINING
HIGH LEVELS OF FISH OIL**

A thesis presented in partial fulfilment of
the requirement for the degree of
Master of Nutritional Sciences
at Massey University, Palmerston North,
New Zealand

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1999

LIST OF ABBREVIATIONS

α -TO \cdot	α -Tocopherol radical
α -TOH	α -Tocopherol
$^1\text{O}_2$	Active oxygen
$^3\text{C}\cdot$	Triplet excited carotenoid
^3H -TdR	Tritiated thymidine
$^3\text{O}_2$	State oxygen
AAFCO	Association of America Feed Control Officials
Ascorbate \cdot	Ascorbate radical
BrdU	5-Bromo-2'- deoxyuridine
Caro	Carotenoids
CAT	Catalase
Con A	Concanavalin A
CPM	Counts per minute
ELISA	Enzyme-linked immunosorbent assay
FRAP	The ferric reducing ability of plasma
GSH	Reduced glutathione
GSHPx	Glutathione peroxidase
GSH _{red}	Glutathione reductase
GSSG	Oxidised glutathione
H ₂ O ₂	Hydrogen peroxide
HO \cdot	Hydroxyl radical
LO ₂ \cdot	Lipid hydroperoxide radicals
LOOH	Lipid hydroperoxides
LPO	Lipid peroxides
MCDP	10-N-Methylcarbamoyl-3,7-dimethylamino-10 H-phenothiazine
NADP	Triphosphopyridine nucleotide
NO ₂ \cdot	Nitrogen dioxide
NRC	National Research Council
O ₂ \cdot^-	Superoxide anion
OH \cdot	Hydroxyl radical
PA ₂	Phospholipase A ₂
PHA	Phytohemagglutinin
PHGSHPx	Phospholipid hydroperoxides glutathione peroxidase
PUFA	Polyunsaturated fatty acids
PWM	Pokeweed mitogen
R \cdot	Carbon-centered radical
RBC	Red blood cell
ROO \cdot	Fatty acid hydroperoxide radicals
ROOH	Fatty acid hydroperoxides
Se	Selenium
SI	Stimulation index
SOD	Superoxide dismutase
TBA	Thiobarbituric acid

ABSTRACT

The vitamin E (α -tocopherol) requirement of adult cats fed diets containing high levels of fish oil was investigated. Thirty-two (16 male, 16 female) adult domestic cats (*Felis catus*) were randomly allocated to four groups according to sex and fed one of four experimental diets (A, B, C, and D) for 126 days. The cats were housed in large outdoor pens in groups of 8 cats. Diets A, B, C and D contained approximately 300 g of fish oil per kg diet dry matter and were supplemented to contain 0, 5, 10, and 15 IU DL- α -tocopheryl acetate per g added fish oil per kg diet, respectively. The diets were provided *ad libitum* with water being available at all times. Food intake was measured daily and body weights were measured at weekly intervals. Blood samples were taken from the jugular vein of each cat at bi-weekly intervals during the study. Blood samples were analysed for plasma α -tocopherol, red blood cell H_2O_2 (4 and 2 %) haemolysis, the ferric reducing ability of plasma, plasma lipid peroxides, plasma triglycerides, alkaline phosphatase and whole blood lymphocyte proliferation.

All cats remained healthy throughout the study except one female cat who was removed after 3 weeks due to poor food intake. The four diets were analysed and found to be free of peroxides. The average daily metabolisable energy intake of the cats on diet A, B, C and D at the end of study were similar and were 289, 261, 256, and 267 $\text{kJ}\cdot\text{kg}^{-1}$ body weight, respectively. No clinical signs of vitamin E deficiency were observed in any of the cats. The plasma α -tocopherol concentrations of the cats in the four groups at the start of the study were not significantly different between the four groups (mean \pm SEM, $3.4 \pm 0.2 \mu\text{g}\cdot\text{ml}^{-1}$). When the cats were fed diet A (unsupplemented), the mean plasma α -tocopherol concentration remained relatively low and the RBC 4 % H_2O_2 haemolysis remained high, while the RBC 2 % H_2O_2 haemolysis decreased consistently. Plasma lipid peroxides remained relatively low throughout the study. The ferric reducing ability of plasma status was compromised in the cats on the unsupplemented diet. There was no significant ($P < 0.05$) difference in any of the response parameters measured amongst the cats fed diets B, C and D except for the RBC 4 % H_2O_2 haemolysis of the cats on diet B which was significantly higher than those on diet C and D at week 4 and week 8, and the LPO value of the cats on diet D which was significantly higher than those of the cats on diet B and C at week 4.

The vitamin E requirement of adult cats fed a high level of fish oil, using the response parameters measured, was estimated to be between 0 and 5 IU of vitamin E per g added fish oil per kg diet. The current recommendation of the Association of American Feed Control Officials (10 IU vitamin E/g fish oil/kg diet) appears to be well in excess. The results from the present study also showed that there was no beneficial effect of dietary vitamin E on whole blood cell proliferation when vitamin E levels were 150 % of the recommendations of the Association of American Feed Control Officials. The vitamin E requirement of adult cats to optimise immune response warrants further investigation.

ACKNOWLEDGEMENTS

I would like to sincerely thank to my chief supervisor, Dr Wouter Hendriks for his advice, patient and guidance during the course of this study. I extend my thanks to my second supervisor, Associate Professor Brian Jordan for his corrections and valuable comments.

I would also like to thank Dr Kay Rutherford for her laboratory supervision, Mr Shane Rutherford, Ms Maggie Zou, Dr John McIntosh, Dr Philip Pearce for their expert assistance in the chemical analysis of α -tocopherol, fatty acids and the ferric reducing ability of plasma and Ms Rosemary Watson for performing the red blood cell hydrogen peroxide haemolysis assay.

I would like to thank the following people for their contributions to this work:

Mrs Linley Fray for her technical assistance with the cell proliferation assay.

Mrs Heather Nicol, Ms Karin Weidgraaf for their technical assistance in the conduct of the study.

Drs Tsegaw Belay and Mark Newcome of Heinz Pet Products, Terminal Island, Los Angeles, California, USA for manufacturing the experimental diets.

I acknowledge the support and encouragement of Mr Terry McGrath, Mr Parackal Mathew and Mrs Sherly Mathew during my study.

I am extremely thankful to my wife for her patience and support throughout the study. I extend my gratitude to my parents for fostering my education, which allowed me to undertake this study. I thank my parents-in-law for their support, and encouragement throughout this study.

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GENERAL INTRODUCTION

The domestic cat (*Felis catus*) is a member of the Felidae family of the order Carnivore and one of the most popular companion animals. Part of their attraction lays in their playful behaviour (Houpt *et al.*, 1988). In recent years researchers have discovered that the relationship between humans and their pets provides numerous physiological and psychological benefits to the owner (Case *et al.*, 1995).

Besides proper health care and medical attention, nutrition is considered to be an important component of the care of cats. Nutritional balance and preferences of diets must be considered when a diet is formulated for cats by an animal nutritionist (Case *et al.*, 1995). It is well known that many cats prefer fish and consequently, numerous cat foods are composed of fish or flavoured with fish (Houpt *et al.*, 1988). However, there have been several reports of vitamin E deficiency in cats as a result of the exclusive feeding of fish and fish based diets (Cordy and Stillinger, 1953; Coffin and Holzworth, 1954; Munson *et al.*, 1958; Griffiths *et al.*, 1960; Watson *et al.*, 1973; Gaskell *et al.*, 1975; Flecknell and Gruffydd-Jones, 1978; Summers *et al.*, 1982; Koutinas *et al.*, 1993; Tidholm, 1996) over the last 50 years.

The vitamin E requirements of other animal species such as humans, rats, pigs, dogs, and guinea pigs have been extensively studied (Van Vleet, 1975; Farrell *et al.*, 1985; Hakkarainen *et al.*, 1986; Jensen *et al.*, 1988a; Mahan, 1991; Meydani *et al.*, 1991; Cho and Choi, 1994; Barja *et al.*, 1996; Wang *et al.*, 1996; Kubo *et al.*, 1997). These studies have demonstrated that the *in vivo* vitamin E requirements are markedly influenced by dietary composition. A high dietary level of polyunsaturated fatty acids increases the requirement for vitamin E as a result of the increased susceptibility of tissues to peroxidation (Duthie, 1993). The dietary vitamin E requirement of cats has been set at 30 IU·kg⁻¹ dry matter: a figure mostly extrapolated from other animal species (NRC, 1986). In order to prevent vitamin E deficiency in cats fed commercially sold, fish based diets, the Association of America Feed Control Officials (AAFCO, 1997) recommends that diets containing fish oil should be supplemented with 10 IU of vitamin E for every g of fish oil per kg diet. AAFCO (1997) failed to provide evidence to substantiate this value and, therefore, the exact vitamin E requirements of cats fed high levels of polyunsaturated fatty acids are still largely unknown.

The main objective of this study was to determine the vitamin E requirement of cats fed high dietary levels of polyunsaturated fatty acids from fish oil. This study was also undertaken to obtain baseline data on α -tocopherol levels in blood plasma of adult cats, which can be used in the diagnosis of vitamin E deficiency.